

**European Patent No 1 096 959 B1**

**(European Application No 99928964.8)**

**Life Science TGO, S.R.L.**

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**Opposition by Novozymes Biologicals, Inc**

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## **REQUESTS**

1. We request revocation of the Patent in its entirety for all contracting states on the grounds that the claimed invention lacks novelty (Article 54 EPC; Article 100(a) EPC); lacks inventive step (Article 56 EPC; Article 100(a) EPC); and lacks sufficiency of disclosure (Article 83 EPC; Article 100(b) EPC).
2. In the event that the Opposition Division (OD) is unable to grant this request based on the written submissions, we request oral proceedings pursuant to Article 116 EPC.

## **LACK OF NOVELTY**

3. The claimed invention was made available to the public before the priority date of the Patent by (1) the sale of compositions comprising spores of *Bacillus licheniformis*, *Bacillus subtilis* and *Bacillus polymyxa*, and compositions comprising spores of *Bacillus licheniformis*, *Bacillus pasteurii*, *Bacillus laevolacticus* and *Bacillus amyloliquefaciens* for deodorising carpets; (2) the public distribution of technical sales literature relating to (1); and (3) by the sale of OUTRIGHT as acknowledged on page 3, lines 6 to 9 of the Patent (although the characterisation of the properties of OUTRIGHT in the Patent is wrong). We refer the OD to the statements of Ms Lois Davis (D1) and Mr Domenic A Paone (D2) and the Exhibits attached thereto.

### ***Claim 1***

4. Claim 1 is directed at a method for controlling odour associated with deposits of organic material on a carpet by applying to the carpet a preparation of dormant bacteria which, when activated, are effective to control odours.
5. In the only exemplified embodiment of Claim 1 the dormant bacteria are a mixture of spores of *Bacillus licheniformis*, *Bacillus pasteurii*, *Bacillus laevolacticus* and *Bacillus amyloliquefaciens*.
6. We refer to D1 and D2. Exhibits B and C of D1 and D2 are the same technical sales information sheets widely distributed to customers and potential customers soon after their production (January 1997 in the case of Exhibit B and June 1997 in the case of Exhibit C) and which describe two products produced by Sybron

Biochemical called Bi-Chem MSB 4X (Exhibit B) and Bi-Chem BioClean (Exhibit C). See paragraphs 5 to 7 of D1.

7. Exhibit B shows that Bi-Chem MSB 4X contains a blend of four *Bacillus* spores (see under "Product Characteristics" on page 2) for use on carpets (see under "Applications" on page 1). The *Bacillus* spores germinate in the presence of organic material (see "Superior Germination and Outgrowth" on page 2) and enzymatically degrade the waste (see "Accelerated Enzymatic Degradation" and "General Organic Waste Degradation" on page 2). Since organic waste is known to cause odours, the use of Bi-Chem MSB 4X on carpets as instructed in Exhibit A will lead to the control of odours caused by organic waste.
8. Thus, Exhibit B of D1 and D2 destroys the novelty of Claim 1.
9. Exhibit C shows that Bi-Chem BioClean contains a blend of *Bacillus* spores (see "Product Characteristics" on page 2) for use on carpets (see, for example, title on page 1 "Effective Cleaning and Degradation of Carpet Trapped Organics"). It is specifically stated that Bi-Chem BioClean has the "*ability to degrade organics in carpet fibers*" and that "[t]hese organic spills lead to stains and odors" and that "*BI-CHEM BIOCLEAN effectively degrades these organics resulting in odor control and stain removal*" (see paragraph spanning columns 1 and 2 of page 3).
10. The graph on page 3 indicates germination and growth of spores on a carpet with spilled organics. (As an aside, it is useful to compare this graph with the graph in Figure 4 of the Patent. They are substantially identical.)
11. Thus, Exhibit C of D1 and D2 destroys the novelty of Claim 1.
12. The novelty of Claim 1 is also destroyed by the sale of Bi-Chem BioClean to Qualitech Systems, Inc of New Hyde Park, NY, USA in April 1998 as demonstrated in paragraph 10 of D1 and by reference to the sales invoice (Exhibit I of D1). The detailed composition of Bi-Chem BioClean is described in paragraph 7 of D2 and Exhibit B of D1 and D2 shows that a principle use of Bi-Chem BioClean is for degrading trapped organics in carpets in order to deodorise the carpet.
13. The novelty of Claim 1 is also destroyed by the sale of Bi-Chem MSB 10X to Carpet Plus before April 1997 (see paragraph 11 of D1 and Exhibit J; see also paragraph 7 of D2 which indicates that Bi-Chem MSB 10X contains *Bacillus* spores).

14. The novelty of Claim 1 is also destroyed by the sale of Bi-Chem MSB 20XNF to RPG Manufacturing before April 1997 (see paragraph 17 of D1 and Exhibit M; see also D2 at paragraph 7).
15. We refer again to D1 and D2. Exhibit A of both declarations are the same technical sales information sheets relating to a further product produced by Sybron Biochemicals, namely Bi-Chem GC 600L. As explained in D2 at paragraph 6, Bi-Chem GC 600L is a composition comprising spores of *Bacillus licheniformis*, *Bacillus subtilis* and *Bacillus polymyxa* sold for use in carpet care. As explained in D1 at paragraphs 4 and 7 the information sheets relating to this product (Exhibit A of both D1 and D2) were widely distributed to customers and potential customers shortly after November 1994. Pages 7 and 9 of Exhibit A note that the product is a synergistic, three strain blend of stable *Bacillus* spore-forming microorganisms.
16. Thus, Exhibit A of D1 and D2 destroys the novelty of Claim 1.
17. The novelty of Claim 1 is also destroyed by the sale of Bi-Chem GC 600L to numerous customers before July 1998 as set out in paragraph 8 of D1. In particular, Bi-Chem GC 600L was sold to Cobb Carpet Supply, Dallas, Texas in February and May 1991, to Rug Doctor in Fresno, California in August 1992 and November 1994, and to Alken Murray Corp of New York in September 1994. In each case, the sale was for use in deodorising carpets as set out in the information sheets.
18. We refer to paragraph 12 and Exhibit K of D1 which is a letter from Cobb Carpet Company confirming the receipt and use of Bi-Chem GC 600L as early as September 1993. The product was used in Cobb Carpet's Dynachem Enzyme deodorant for application on carpets and sold in the southern region of the United States.
19. We refer to paragraph 13 and Exhibit L of D1 which is a letter from Spartan Chemical Company acknowledging that the Bi-Chem GC 600L product was used commercially in a Spartan product since April 1992.
20. The novelty of Claim 1 is also destroyed by the sale of OUTRIGHT. The Patent at page 3, lines 6 to 9 correctly acknowledges that The Bramton Company of Dallas sold OUTRIGHT before the date of the Patent for controlling odour in carpets. Contrary to the assertion in the Patent that OUTRIGHT is a preparation of active *Bacillus* in a vegetative form, it is in fact a preparation of *Bacillus* spores sold to

The Bramton Company by Sybron Biochemical (see paragraph 14 of D2 and paragraph 17 of D1).

21. All the spore products discussed above are activated (ie the spores germinate) in the presence of organics (see D2 at paragraph 9).
22. In all of the sales, D1 at paragraphs 8 and 10 confirms that technical sales information appropriate for the product was despatched to the purchasers of the products.
23. Thus, D1 and D2 provide evidence that the method of Claim 1 was made available to the public before the priority date of the Patent.

#### **Claim 2**

24. Claim 2 requires the dormant bacteria to be "sporulated forms of one or more strains selected from the bacterial genera (*sic*) *Bacillus*". Thus, Claim 2 lacks novelty for all the same reasons as Claim 1 since in all cases *Bacillus* spores are used.

#### **Claim 3**

25. Claim 3 requires the dormant bacteria to be sporulated forms of one or more strains selected from *Bacillus licheniformis*, *Bacillus pasteurii*, *Bacillus laevolacticus* and *Bacillus amyloliquefaciens*. As discussed in detail in D2, both Bi-Chem MSB 4X (and 10X and 20X) and Bi-Chem BioClean contain a mixture of spores of these four *Bacillus* species (see paragraphs 7 and 8 of D2). Thus, the sale of Bi-Chem BioClean before the priority date of the Patent made the subject matter of Claim 3 available to the public before the priority date and so Claim 3 lacks novelty.
26. Furthermore, the sale of OUTRIGHT before the priority date of the Patent, as acknowledged by the Patentee, destroys the novelty of Claim 3 since OUTRIGHT contains spores of *B. licheniformis*, *B. pasteurii*, *B. laevolacticus* and *B. amyloliquefaciens* (see paragraph 14 of D2).
27. Similarly, the sale of Bi-Chem GC 600L (which contains a mixture of spores including those of *Bacillus licheniformis*; see paragraph 6 of D2) before the priority date made the subject matter of Claim 3 available to the public before the priority date.

28. Although not prior art, we refer the OD to WO 99/46350 (D3; also it is Exhibit E of D2). This is a patent application by Sybron which relates to a deodorizing composition for carpet care containing spores of the four *Bacillus* spp (see page 9, line 20 and Table 4 on page 10; also, see paragraphs 7 and 12 of D2) but in this case the invention relates to including a microbial activity enhancing component (such as yucca schidigera) or an odour neutralizing component (such as propylene carbonate) in the known mixture of four *Bacillus* spp.
29. WO 97/25865 (D4; also it is Exhibit D of D2) is a patent application by Sybron which demonstrates that spores of all of the *Bacillus* spp mentioned in the Patent were being used by Sybron before the priority date (see, for example, "Microorganism" on page 5 *et seq*) and that they were being used for cleaning and desanitizing (see also paragraphs 6 and 11 of D2).

#### ***Claims 4 and 5***

30. As indicated in paragraph 12 of D2, Bi-Chem BioClean has a spore count of  $5.4 \times 10^7$  per ml. Mr Paone states that around 3-5 ml of this would be used per square inch of carpet which we submit falls within the concentration range of Claims 4 and 5.

#### ***Claim 6***

31. Bi-Chem BioClean contains a mixture of *Bacillus* spores in the following ratios:

licheniformis 70 : pasteurii 10 : laevolacticus 10 : amyloliquefaciens 10

(see D2 at paragraph 7).

32. Thus, the subject matter of Claim 6 was made available to the public before the priority date of the Patent.

#### ***Claim 22***

33. Claim 22 is directed at the inevitable result of carrying out the method of Claim 1 on a carpet and so lacks novelty for the same reasons as Claim 1.

### ***Claims 23 to 27***

34. Claims 23 to 27 are directed at the inevitable results of carrying out method of Claims 2 to 6, respectively, and so lack novelty for the same reasons as Claims 2 to 6.

### **LACK OF INVENTIVE STEP**

#### ***Claim 1***

35. Paragraph [0007] acknowledges that various *Bacillus spp* are useful for odour control in various settings and notes, in particular, that preparations of “*active Bacillus in a vegetative form suitable for spraying or otherwise distributing on a deposit, especially of pet urine and feces, on a carpet for controlling odor are presently marketed by The Bramton Company of Dallas, Texas under the trademark OUTRIGHT*”.
36. We have provided evidence that the prior sale of OUTRIGHT destroys the novelty of Claim 1. However, if the OD is unwilling to accept this, Claim 1 lacks an inventive step over the Patentee’s admissions in relation to OUTRIGHT for the following reasons.
37. It is well known that *Bacillus spp* are able to form spores under conditions of nutrient limitation, and that these spores will germinate in the presence of moisture and nutrients for example, see D5.
38. There is nothing inventive in replacing the vegetative form of *Bacillus* with spores which are known to be able to germinate into the vegetative form under conditions where there will be moisture and nutrients (eg presence of urine).
39. Claim 1 lacks an inventive step for this reason alone.
40. In the event that the OD does not consider Exhibits A, B and C novelty destroying for Claim 1, they destroy the inventive step since they clearly suggest the use of *Bacillus* spores to deodorise carpets in the way claimed.
41. Claim 1 also lacks an inventive step over WO 97/25865 (D4) alone or in combination with any of Exhibits A, B or C, or the known sale of OUTRIGHT. D4 describes compositions of spores (page 3, line 1) of various *Bacillus spp* (see “Microorganisms” on page 5 *et seq*) for use in cleaning and sanitizing. It is noted that the *Bacillus spp*



have excellent waste-degrading capabilities (page 5, line 22). There is nothing inventive in using such known compositions to degrade waste on a carpet. This is particularly so in light of the disclosures in Exhibits A to C and in light of the acknowledged sale of OUTRIGHT.

### **Claim 2**

42. Claim 2 lacks an inventive step for the same reason as Claim 1.

### **Claim 3**

43. *B. licheniformis*, *B. pasteurii*, *B. laevolacticus* and *B. amyloliquefaciens* are well known, non-pathogenic species of *Bacillus* which are well known to produce enzymes which are able to digest proteins, carbohydrates, lipids and the like, and are able to germinate under suitable conditions. This is acknowledged in the Patent on page 3, lines 56 to page 4, line 3 where it is noted, for example, that *B. pasteurii* is known for superior lipase production, while *B. laevolacticus* has a very fast germination cycle, and *B. amyloliquefaciens* is high in production of protease enzymes.
44. Thus, these *Bacillus spp* are the obvious choice since, for example, D4 indicates that they have desirable waste-degrading characteristics. Also, they are used in OUTRIGHT and so Claim 3 lacks an inventive step for this reason and the reasons given for Claims 1 and 2.

### **Claims 4 and 5**

45. There is nothing inventive in the selection of the claimed concentration of "cells" to use in treating the carpet. The concentrations given are readily arrived at by the skilled person in carrying out the method of Claim 1 and no unexpected results are demonstrated in the Patent with the use of these concentrations.
46. In addition, a concentration range of  $10^6$ - $10^8$  spores per ml is given in Table 4 of D4 and it is reasonable to use around 1 ml of spore solution per g of carpet fibre (see, for example, paragraph 13 of D2).
47. Thus, Claims 4 and 5 lack an inventive step.

### ***Claims 6 and 7***

48. There is nothing inventive in the selection of the particular claimed ratios of the particular *Bacillus* spp since it is obvious to make the selection based on the known attributes of the different species (eg their lipase activity, protease activity etc).
49. Furthermore, no unexpected results are demonstrated in the Patent with the use of the particular ratios of the particular species.

### ***Claims 8 to 11***

50. There is nothing inventive in using a stain blocking chemical in a method controlling odour in carpets caused by organic material which may also cause stains.
51. Stain-blocking chemicals for treating carpets are known from, for example, US Patent Nos 4,680,212 (D6) and 4,925,707 (D7). The specific chemicals mentioned in Claim 9 of the Patent are common stain-blocking chemicals and are described in D7 (see, for example, Claim 26) and in D6 (see, for example, Claims 10, 11 and 12).
52. There is nothing inventive in the treatment rates mentioned in Claims 9 and 10. In particular, D6 column 5, lines 14 and 15 indicate that a preferred treatment rate is 0.35% by weight.

### ***Claim 12***

53. There is nothing inventive in using one or more anti-soil fluorochemicals in a method of controlling odour in a carpet associated with organic waste since such compounds are well known as anti-soil treatments for carpets. The Patent at page 5, lines 1 and 2 refers to well-known fluorochemicals for treating carpets, and these are used in combination with stain-blocking chemicals in D7 (for example, see Example I, column 9, lines 11 to 16 where TEFLON is used).

### ***Claim 13***

54. As discussed in relation to the novelty of Claim 1, the use of dormant bacteria in an aqueous composition for controlling odours, for example in carpets, is known. Such a composition is also lacking in inventive step as discussed above.

55. There is nothing inventive in including in such a composition one or more stain-blocker chemicals which were well known for use in treating carpets (see comments relating to Claims 8 to 11 above).

***Claim 14***

56. Claim 14 lacks an inventive step for the same reason as Claim 13. In particular, as discussed in relation to Claims 1 to 3, spores of *Bacillus spp* are known for controlling odour. None of the other genera of bacteria are inventive since they are known to produce enzymes capable of breaking down organic material and these bacteria have been used for deodorising (see page 3, lines 50 to 54 of the Patent). To the extent that the OD may consider that the use of some bacterial species or strains within these genera are inventive, there is not a sufficient disclosure of which ones or how to make them dormant (if this is, indeed, possible) or how to use them to give the claimed effect.

***Claim 15***

57. The particular bacterial species are those present in known compositions for controlling odour in carpets as discussed in relation to Claims 1 to 3. Thus, Claim 15 lacks an inventive step.

***Claims 16 and 17***

58. Claims 16 and 17 lack an inventive step for the reasons given for Claims 4, 5 and 14.

***Claims 18 and 19***

59. Claims 18 and 19 lack an inventive step for the reasons given for Claims 6, 7 and 14.

***Claim 20***

60. Claim 20 lacks an inventive step for the reasons given for Claims 8 to 11 and 14.

### ***Claim 21***

61. Claim 21 lacks an inventive step for the reasons given for Claims 12 and 14.

### ***Claims 22 to 33***

62. Use of the known method of Claims 1 to 3 results in the production of a carpet according to Claims 22 to 24. Thus, even if the OD does not consider Claims 22 to 24 to lack novelty they undoubtedly lack inventive step for the same reasons as Claims 1 to 3.
63. The features of the carpet set out in Claims 25 to 33 are all obvious from using the claimed methods or compositions as discussed above.
64. Thus, none of Claims 22 to 33 have an inventive step.

### **LACK OF SUFFICIENCY**

***The only preparation of dormant bacteria shown to work is a specific combination of spores of Bacillus licheniformis, Bacillus pasteurii, Bacillus laevolacticus and Bacillus amyloliquefaciens***

65. The independent claims (Claims 1, 13, and 22) refer to “a preparation of dormant bacteria” for controlling odour associated with deposits of organic material which can cause odours in carpet.
66. Apparently, this preparation can be of *any* dormant bacteria which, when activated, are effective to control odors. In contrast to the broad scope of these claims, the *only* example of a preparation of dormant bacteria described in the Patent is a *specific combination* of spores of *B. licheniformis*, *B. pasteurii*, *B. laevolacticus* and *B. amyloliquefaciens*.
67. It is well established EPO jurisprudence that the protection conferred by a patent should correspond to the technical contribution to the art made by the disclosure of the invention described therein, which excludes the patent monopoly being extended to subject matter which, after reading the patent specification, would still not be at the disposal of the skilled person (see, for example, T409/91 and T435/91). To the extent that there is any contribution to the art (which we dispute is the case anyway), it is restricted to the particular combination of *Bacillus* spores since finding any further

preparations of dormant bacteria which have the claimed effect would place an undue burden on the skilled person. No guidance is given in the Patent as to which other bacterial strains or species have the desired properties or how to prepare dormant forms of them. We acknowledge that there is a reference to various genera of bacteria, for example on page 3, lines 51 to 54, but this amounts to merely an acknowledgement that certain species and strains within these genera are known for use in degrading organic material and for deodorising. There is no disclosure of which ones, if any, can be made dormant and would be effective in the method claimed.

### **DOCUMENTS CITED**

1. Declaration of Ms Lois Davis and Exhibits A to M attached thereto.
2. Declaration of Mr Domenic A Paone and Exhibits A to E attached thereto.
3. WO 99/46350.
4. WO 97/25865.
5. Extract from "General Microbiology" by Schlegel, H G, Seventh Edition, Cambridge University Press, page 83.
6. US Patent No 4,680,212.
7. US Patent No 4,925,707

IN THE MATTER of the  
opposition to EP 1 096 959 B1 to  
Life Science TGO, S.R.L. by  
Novozymes Biologicals, Inc at  
the European Patent Office

### DECLARATION

1. I, Lois Davis, am Global Business Manager at Novozymes Biologicals, Inc ("Novozymes"), 5400 Corporate Circle, Salem, VA 24153, USA and have been since 2002. I am responsible for the institutional and household business, a business I have been involved in since 1991.
2. Novozymes acquired the business of Sybron Biochemical ("Sybron") in 2001, including their Bi-Chem products, and before the acquisition I was employed by Sybron as marketing manager for the institutional and household, and industrial waste treatment businesses.
3. Sybron, and subsequently Novozymes, have an ongoing business relating to the production and sale of blends of multiple *Bacillus* spores for various applications including carpet and fabric care as well as for drain line and grease trap treatment, as a bathroom cleaner and deodorizer, as a laundry pre-spotter, for portable toilet treatment and for septic and waste treatment.
4. I refer to Exhibit A which is a collection of technical sales information sheets and other information for the Sybron product, Bi-Chem GC 600L. These information sheets were prepared under the supervision of my predecessor, Doug Dent, by a former employee, Lisa Marie D'Anzi, in 1994 as indicated on page 3 by the notation "LMD 6/94", on page 4 by the notation "LMD 11/94", on page 6 by the notation "10/94-LMLD" and on page 8 by the notation "10/94-LMLD". This notation on the various information sheets indicates the date that they were prepared or revised.
5. I refer to Exhibit B which is a technical sales information sheet for the Sybron product called "Bi-Chem MSB 4X". This information sheet was prepared under the supervision of my predecessor, Doug Dent, by Lisa D'Anzi in January 1997 as indicated on the second page by the notation "1/97-LMLD".
6. I refer to Exhibit C which is technical sales information (product summary and technical data sheets) for the Sybron product called

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"Bi-Chem Bioclean". These information sheets were also prepared by Lisa D'Anzi under the supervision of my predecessor, Doug Dent, but this time in June 1997 as indicated by the notation "6/97-LMLD" on pages 2 and 4.

7. The information sheets in Exhibits A, B and C were distributed freely to customers and potential customers (including virtually all significant janitorial supply companies), soon after they were prepared or revised.
8. I can also confirm that prior to July 1998 Sybron had sold preparations of *Bacillus* spores for deodorising carpets to at least the following carpet cleaning companies: Alken Murray Corp, Cobb Carpet, Stanley Steemer, Rug Doctor, Bridgepoint, Carpet Plus - Denver, Spartan, RepZ, Betco, Kleemrite, State Industrial and Nilodor and I confirm that the information sheets appropriate to the particular product purchased were sent to these customers.
9. For example, Bi-Chem GC 600L was sold to Cobb Carpet Supply of Dallas, Texas in February and May 1991 as shown by the invoices attached as Exhibits D and E (shipment dates of 2/04/91 and 5/13/91). It was also sold to Rug Doctor in Fresno, California in August 1992 and again in November 1994 as shown by the invoices attached as Exhibits F and G (shipment dates 8/06/92 and 11/18/94). It was also sold to Alken Murray Corp of New York in September 1994 as shown by the invoice attached as Exhibit H (shipment date 9/14/94). I believe that in all instances the Bi-Chem GC 600L product was sold for use in carpet cleaning and deodorising as suggested on the information sheets for the product as set out in Exhibit A.
10. Bi-Chem Bioclean was sold to Qualitech Systems, Inc of New Hyde Park, NY in April 1998 as shown in Exhibit I which is an invoice dated 9 April 1998 (4/09/98) showing a shipment date of 8 April 1998 (4/08/98). I believe that the Bi-Chem Bioclean product was sold for use in carpet cleaning and deodorising as suggested in the information sheets for the product as set out in Exhibit C. I confirm that the information sheets for this product were sent to the customer.
11. Bi-Chem MSB 10 X, another product containing the same four *Bacillus* spores as MSB 4X, was sold to Carpet Plus as least as early as April 1997 as indicated in Exhibit J which is a sales summary sheet produced on 30 April 1997.

*Handwritten signature*

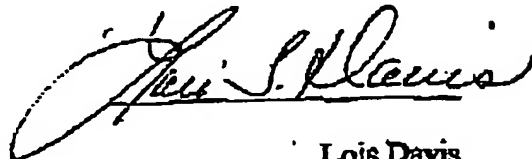
12. I refer to Exhibit K. Exhibit K is a letter that I received on 25 May 2004 from Mr Lawrence Cobb of Cobb Carpet Supply. As I have stated in paragraph 9 above, Bi-Chem GC 600L was sold to Cobb Carpet Supply in 1991. Mr Cobb's letter confirms that his firm used this product in their Dynachem Enzyme Deodorant for application on carpets which was sold in the southern region of the United States. Mr Lawrence explains in his letter that he has dated formulation records of GC-600L 10X being used as of September 1993.
13. I refer to Exhibit L. This is a letter that I received on 26 May 2004 from Mr William Schallitz, Vice President, Research & Development at Spartan Chemical Company, Inc indicating that the Bi-Chem GC 600L product was used as the basis of the Spartan Consume product which was commercialised in April 1992 and has always been labelled with the directional heading of "Carpet Odor and Stain Remover".
14. As discussed in more detail by my colleague, Domenic Paone in his accompanying declaration which I have read and agree with, Bi-Chem GC 600L contains a mixture of spores from *Bacillus licheniformis*, *Bacillus subtilis* and *Bacillus polymyxa*. Exhibit A of my declaration is the same as Exhibit A of Domenic Paone's declaration.
15. As discussed in more detail in Domenic Paone's declaration, the products Bi-Chem MSB 4X (and 10X and 20X) and Bi-Chem BioClean contain a blend of spores from *Bacillus licheniformis*, *Bacillus amyloliquefaciens*, *Bacillus pasteurii* and *Bacillus laevolacticus*, present in a ratio of 70:10:10:10, although I do not believe that specific ratios are critical for their use. In particular, the Bi-Chem BioClean preparation sold to Qualitech Systems, Inc contained spores from these four *Bacillus* spp.
16. Exhibits B and C of my declaration are the same as Exhibits B and C of Domenic Paone's declaration.
17. As discussed in the accompanying declaration of Domenic Paone, The Bramton Company of Dallas, Texas has for many years sold, and certainly sold before July 1998, a product for controlling odor on a carpet under the brand OUTRIGHT. The Bramton Company is also known as RPG Manufacturing, and our records show that Sybron sold them Bi-Chem MSB 20 X NF, a product containing the same four *Bacillus* spores as in the Bi-Chem MSB 4X product, before April 1997 as shown in Exhibit M. Exhibit M is a sales summary sheet produced on 30 April 1997. Bi-Chem MSB 20 X NF



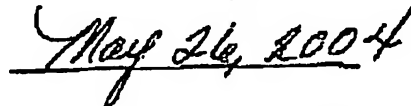
was sold to RPG Manufacturing for the purposes of preparing a formulation for carpet cleaning and odor control; to the best of my knowledge and belief this is the product known as OUTRIGHT.

18. Exhibits D to J and M have been redacted to remove irrelevant price information.

I hereby declare that the facts set out above are true to the best of my knowledge and belief



Lois Davis



Date

# **Declaration by Lois Davis**

## **Exhibit A**

# BI-CHEM<sup>™</sup> BACTERIAL CULTURES. THE LITTLE GUYS WHO CAN HELP YOU MAKE IT BIG.



Sybron Biochemical offers you a complete line of scientifically developed Bioformulations in liquid or dry form. These unique blends of selected and patented Bi-Chem bacteria will solve your customers' special problems with grease digestion (TD Series), odor control (GC Series) and waste degradation (SM Series). And they're all available fully formulated or as concentrates.

## TD SERIES

Bi-Chem TD Series bacteria are specifically designed for rapid grease liquification and digestion in traps, drains and municipal collection systems. TD contains a blend of patented biostains that digests fat, grease and oil ten times faster than competitive

products. TD is available in liquid or dry formulations.

## GC SERIES

Bi-Chem GC Series includes GC 600L, an all-purpose odor controller and waste degrader in liquid form. GC 600L provides exceptional market appeal with its pleasant fragrance and attractive appearance. Best of all, GC destroys odors and stains at their source, instead of masking them.

Bi-Chem SM Series cultures are

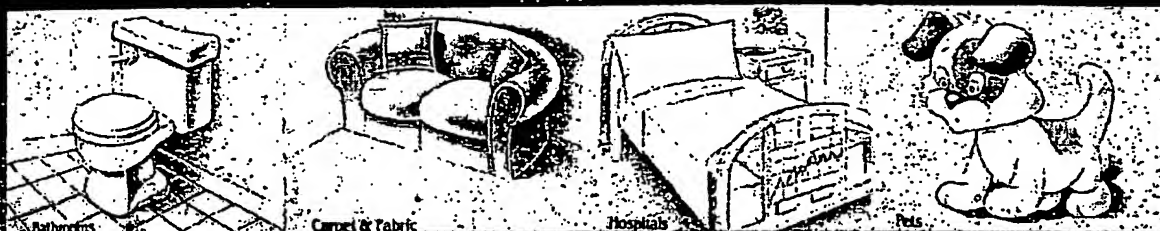
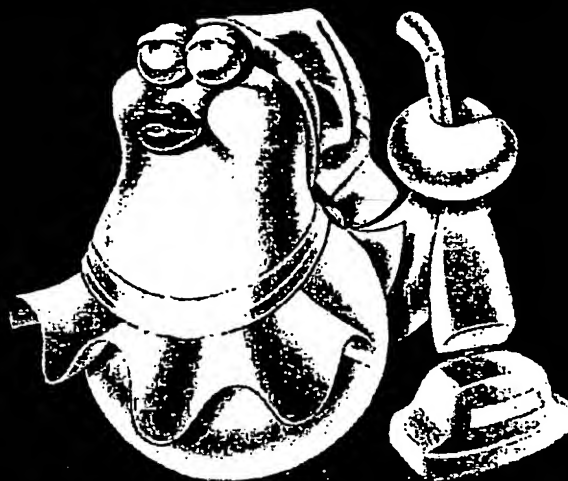
designed for rapid liquification and digestion of difficult waste compounds such as detergents, fats, oils, greases and cellulose. These cultures provide the best protection your customers can buy for waste systems and significantly improve the performance of septic tanks, lagoons, activated sludge and waste holding basins. SM is available in liquid or dry formulations.

For more information on how you can make it big with our little guys—TD Series, GC Series and SM Series Bacteria—please contact Doug Dent, Manager Sanitary Products, or Mary Houston, Sales Coordinator, at 800/257-9428 or 609/893-1100 in NJ.

SOLVING A WORLD OF PROBLEMS  
 THROUGH BIOTECHNOLOGY



# 71 SNUFF OUT ODORS WITH BI-CHEM® BACTERIA.



The amazing Bi-Chem GC-600L Series of waste degrading, odor destroying, liquid bio-formulations is another in the complete line of Bi-Chem bacterial products designed for institutional and household applications.

The GC-600L Series is a unique broad-spectrum chemical/bacterial blend which combines the effectiveness of selected biostains with the rapid performance of special surfactants and perfumes. The GC-600L Series provides instant odor control, organic removal and quick cleaning. This unique bio-formulation provides a profitable, safe "natural solution" for all types of odor and stain problems (including milk, vomit, feces, urine, blood, coffee, etc.)

GC-600L is a sales leader

marketed as a carpet and fabric cleaner, laundry spotter, urinal and bathroom cleaner and odor controller. Applications also include resolving problems with floor drains, lines and traps.

GC-600L removes odors and stains at the source, instead of just masking the problem—amazing results from an amazing product! The GC Series is available in ready-to-use liquids, concentrates or non-formulated concentrates for blending at your facility. In addition to providing a complete line of superior

bacterial products backed by over 50 years of experience, we offer marketing assistance, training, seminars, literature reviews and competitive product analyses to our customers at no extra charge.

If you want to eliminate odors, stains and organic problems the natural way, and make a nice profit while you're at it, take a closer look at Bi-Chem GC Series bacterial cultures.

For more information on the Bi-Chem GC Series, our grease eating TD Series or our waste degrading SM Series, please contact Doug Dent, Manager Sanitary Products, or Mary Houston, Sales Coordinator, at 800/257-9428 or 609/893-1100 in NJ. Samples and literature are available at your request.

SOLVING A WORLD OF PROBLEMS  
THROUGH BIOTECHNOLOGY





Types of substrates which the enzymes produced by our bacterial strains in BI-CHEM GC 600 L react with:

Amylase is a starch-digesting enzyme. Starch, a polysaccharide, is a polymer of glucose. Starch is a white tasteless, odorless food substance found in corn, potatoes and other starchy vegetables.

Cellulase is a cellulose-digesting enzyme. Cellulose, a polysaccharide, is a glucose polymer in the cell walls of plants used to make paper.

Lipase is a fat-digesting enzyme. Fats are lipids consisting of fatty acids and glycerol. Lipases break fats down into their fatty acid and glycerol components. Each component is then metabolized separately.

Protease is a protein-digesting enzyme. Protein is composed amino acids. Protein is present in milk, cheese, meat, blood and even soil. Protease is contained in some laundry detergents to remove protein stains.

# BLENDING GUIDELINES



BI-CHEM GC 600L comes in the following formulation concentrations:

- BI-CHEM GC 600L 1X (Product code 9361) which is a ready to use product.
- BI-CHEM GC 600L 4X (9534) is a fully formulated concentrate that needs to be diluted 3 parts water to 1 part product.
- BI-CHEM GC 600L 4X NF (9566) is a non-formulated concentrate that needs to be diluted 3 parts water to 1 part product. Also need to formulate fragrance, nonionic surfactant and nonionic dye or opacifier.
- BI-CHEM GC 600L 6X (9589) is a fully formulated concentrate that needs to be diluted 5 parts water to 1 part product.
- BI-CHEM GC 600L 10X NF (9558) is a non formulated concentrate

## INSTRUCTIONS FOR MIXING GC 600L 10X (NF) CONCENTRATE

1. Preclean blending and packaging equipment with 10-20 parts per million (ppm) of liquid chlorine for 15-20 minutes.
2. Use the entire 55 gallon (208 liter) drum of GC 600L (10X). This will produce a 550 gallon (2,080 liter) formulated product.
3. Dilute one part product with approximately nine parts water (city or tap water can be used as long as it is potable) based on percentages of perfume, surfactant, opacifier and other ingredients. The water temperature range should be 50 - 100 °F (10 - 38°C).
4. Premix small portion of surfactants and perfumes to solubilize the perfume.
5. Add the remainder of surfactants and premixed perfume to the batch. Total surfactant is 3% of the batch size and perfume is 0.425% of the batch size.
6. Add opacifier and or color selection. Total opacifier is .25% of the batch size.
7. During formulation adjust pH between 8 to 8.2. Use either phosphoric acid or caustic soda to lower or raise pH.
8. Blend for a minimum of thirty minutes to assure proper spore separation in solution.
9. Maintain slow agitation during the entire process.
10. Do not allow product to stand without agitation during repackaging to drums or small containers at the time of bottling.

SYBRON CHEMICALS INC.

GC600L (10X) nf  
ONE PRODUCT



## FOR THREE MARKETS

Below is find information to help the blender prepare Sybron raw materials for resale into the trap and drain, carpet care and janitorial markets:

### FORMULATIONS

The basic components of any liquid bacterial formulation are a preserved bacterial base, color, surfactant, opacifier and fragrance. Following is a list of ingredients for each particular type of application:

#### 1. application: trap and drain maintenance (drain opener)

Base culture: GC 600L (10X), NON-FORMULATED

Dye: Blue or green (optional)

Surfactant Ethoxylate or nonylphenol

Fragrance: any fragrance will do so long as it is compatible. one suggestion: pine or neutroleum gamma.

Opacifier: gives the liquid a turbid look.

#### 2. application: Carpet and fabric care

Base culture: GC 600 L (10X), NON-FORMULATED

Dye: Cream or no dye. DO NOT USE OTHER COLORS.

Fragrance: any fragrance will do so long as it is compatible. one suggestion: neutroleum gamma.

Opacifier

#### 3. application: JANITORIAL PRODUCTS (HARD-SURFACE cleaning, ODOR CONTROL)

Base culture: GC 600L (10X), NON-FORMULATED

Dye: Blue or green (optional)

Surfactant Ethoxylate or nonylphenol

Fragrance: any fragrance would do so long as it is compatible.  
one suggestion: citrus or pine scent

Opacifier: gives the liquid a turbid look.

#### Alternate approach:

Use GC 600L (6X). This product comes already formulated with surfactant, dye (cream), and fragrance. For orders greater that 550 gallons Sybron can use other colors for an additional charge.

BIOCHEMICAL™ WASTE TREATMENT PRODUCTS  
BIRMINGHAM ROAD, P.O. BOX 66, BIRMINGHAM, NEW JERSEY 08011  
(609) 593-1100 1-(800) 678-0020 FAX: MARKETING (609) 894-8641  
FAX: ACCOUNTING (609) 593-2063 TELEX: WUD: 834446 WUI: 683-1227

## TYPICAL PRODUCT CHARACTERISTICS

### BI-CHEM®GC 600L 10X ENZA-PLUS

Bacteria Count	2000 Billion/gal.
Bacteria Count when diluted to a 1X	200 Billion/gal.

...fat, grease & oil digesting bacteria  
 ...protein digesting bacteria  
 ...starch digesting bacteria  
 ...cellulose digesting bacteria

Appearance	Creamy, white liquid
Odor	Pleasantly perfumed
Specific Gravity	1.00 - 1.025
pH	8.2 - 8.8
Effective pH Range	5.9 - 9.0
Effective Temperature Range	45° - 105°
Standard Packaging	5 and 55 gal. Drums
Stability	2 year

### OPTIMUM CONDITIONS FOR USE

Bacteria in **BI-CHEM GC 600L 10X ENZA-PLUS** perform within a pH range of 5.9 to 9.0 with the optimum near pH 7.5. Temperature affects the activity of the working solution and action increases with rising temperatures up to 105°F. Diminished activity can be expected below 40°F (5°C).

### STORAGE AND HANDLING

Storage	Store in a cool, dry place
Handling	Avoid excessive inhalation. Wash hands thoroughly with warm, soapy water after handling. Avoid eye contact.

### SYBRON CHEMICALS INC. -- BIOCHEMICAL

Sales and Marketing Offices:	Birmingham Road, P.O. Box 66 Birmingham, NJ 08011 (800) 678-0020 or (609) 893-1100 FAX: (609) 894-8641
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Plant and Research Center:	111 Kesler Mill Road Salem, VA 24153
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10/94-LMLD



# PRODUCT DATA

Improved

## BI-CHEM<sup>®</sup> GC 600L 10X ENZA-PLUS

All-purpose Odor Controller and Waste Degradar

### DESCRIPTION

BI-CHEM GC 600L 10X's improved biochemical formulation consists of a synergistic blend of three selected *Bacillus* microorganisms. This improved formulation will have a bacterial count of 2000 billion/gallon. When diluted to a 1X ready to use product, the bacterial count will be 200 billion /gallon.

Of great importance in the development of BI-CHEM GC 600L 10X's bioformulation was the selection of a blend of biostrains specifically designed to promote optimum enzymatic activity for the production of protease, lipase, amylase and cellulase enzymes. As a result, BI-CHEM GC 600L 10X's synergistic formulation provides excellent breakdown of starches, carbohydrates, tissue, fats, oils and grease.

To further enhance the broad spectrum capabilities of the BI-CHEM GC 600L 10X formulation, special biodegradable surfactants, perfumes and opacifiers were selected and combined with our bioformulation creating a unique bacterial/chemical blend with outstanding broad range functions. Our research efforts on BI-CHEM GC 600L 10X have produced an unsurpassed, safe, natural solution for many types of organic waste, odor and stain problems.

### BENEFITS:

- ◆ Instant odor control
- ◆ Organic removal
- ◆ Quick cleaning action

### APPLICATIONS:

- ◆ Bathroom maintenance (surface cleaning and odor control on fixtures and floor drains)
- ◆ Carpet and fabric care (odor and stain removal of milk, vomit, urine, feces, blood, coffee, wine, etc.)
- ◆ Laundry pre-spotter for organic stains (presoak or spot spray)
- ◆ Waste degrader for septic tanks, wastewater systems (ponds, lagoons, etc.)
- ◆ Trap and drain maintenance for fat deposits and odor control

BI-CHEM GC 600L 10X's unique synergistic bacterial/chemical blend offers exceptional market appeal and performance packed in one biochemical formulation.

A Synergistic, Three-Strain Blend of  
Stable *Bacillus* Spore-forming Microorganisms

SOLVING A WORLD OF PROBLEMS  
THROUGH BIOTECHNOLOGY



## TYPICAL PRODUCT CHARACTERISTICS

### BI-CHEM®GC 600L 4X ENZA-PLUS

Bacteria Count	800 Billion/gal.
Bacteria Count when diluted to a 1X	200 Billion/gal.

...fat, grease & oil digesting bacteria  
 ...protein digesting bacteria  
 ...starch digesting bacteria  
 ...cellulose digesting bacteria

Appearance	Creamy, white liquid
Odor	Pleasantly perfumed
Specific Gravity	1.00 - 1.025
pH	8.2 - 8.8
Effective pH Range	5.9 - 9.0
Effective Temperature Range	45° - 105°
Standard Packaging	5 and 55 gal. Drums
Stability	2 year

### OPTIMUM CONDITIONS FOR USE

Bacteria in **BI-CHEM GC 600L 4X ENZA-PLUS** perform within a pH range of 5.9 to 9.0 with the optimum near pH 7.5. Temperature affects the activity of the working solution and action increases with rising temperatures up to 105°F. Diminished activity can be expected below 40°F (5°C).

### STORAGE AND HANDLING

Storage	Store in a cool, dry place
Handling	Avoid excessive inhalation. Wash hands thoroughly with warm, soapy water after handling. Avoid eye contact.

### SYBRON CHEMICALS INC. -- BIOCHEMICAL

Sales and Marketing Offices:	Birmingham Road, P.O. Box 66 Birmingham, NJ 08011 (800) 678-0020 or (609) 893-1100 FAX: (609) 894-8641
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Plant and Research Center:	111 Kesler Mill Road Salem, VA 24153
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10/94-LMLD

# PRODUCT DATA

Improved

## BI-CHEM<sup>®</sup> GC 600L 4X ENZA-PLUS

All-purpose Odor Controller and Waste Degradar

### DESCRIPTION

BI-CHEM GC 600L 4X's improved biochemical formulation consists of a synergistic blend of three selected *Bacillus* microorganisms. This improved formulation will have a bacterial count of 800 billion/gallon. When diluted to a 1X ready to use product, the bacterial count will be 200 billion/gallon.

Of great importance in the development of BI-CHEM GC 600L 4X's bioformulation was the selection of a blend of biostrains specifically designed to promote optimum enzymatic activity for the production of protease, lipase, amylase and cellulase enzymes. As a result, BI-CHEM GC 600L 4X's synergistic formulation provides excellent breakdown of starches, carbohydrates, tissue, fats, oils and grease.

To further enhance the broad spectrum capabilities of the BI-CHEM GC 600L 4X formulation, special biodegradable surfactants, perfumes and opacifiers were selected and combined with our bioformulation creating a unique bacterial/chemical blend with outstanding broad range functions. Our research efforts on BI-CHEM GC 600L 4X have produced an unsurpassed, safe, natural solution for many types of organic waste, odor and stain problems.

### BENEFITS:

- ◆ Instant odor control
- ◆ Organic removal
- ◆ Quick cleaning action

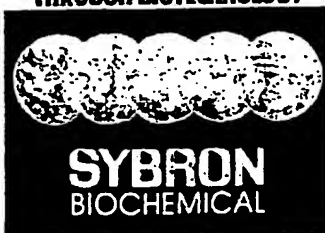
### APPLICATIONS:

- ◆ Bathroom maintenance (surface cleaning and odor control on fixtures and floor drains)
- ◆ Carpet and fabric care (odor and stain removal of milk, vomit, urine, feces, blood, coffee, wine, etc.)
- ◆ Laundry pre-spottér for organic stains (presoak or spot spray)
- ◆ Waste degrader for septic tanks, wastewater systems (ponds, lagoons, etc.)
- ◆ Trap and drain maintenance for fat deposits and odor control

BI-CHEM GC 600L 4X's unique synergistic bacterial/chemical blend offers exceptional market appeal and performance packed in one biochemical formulation.

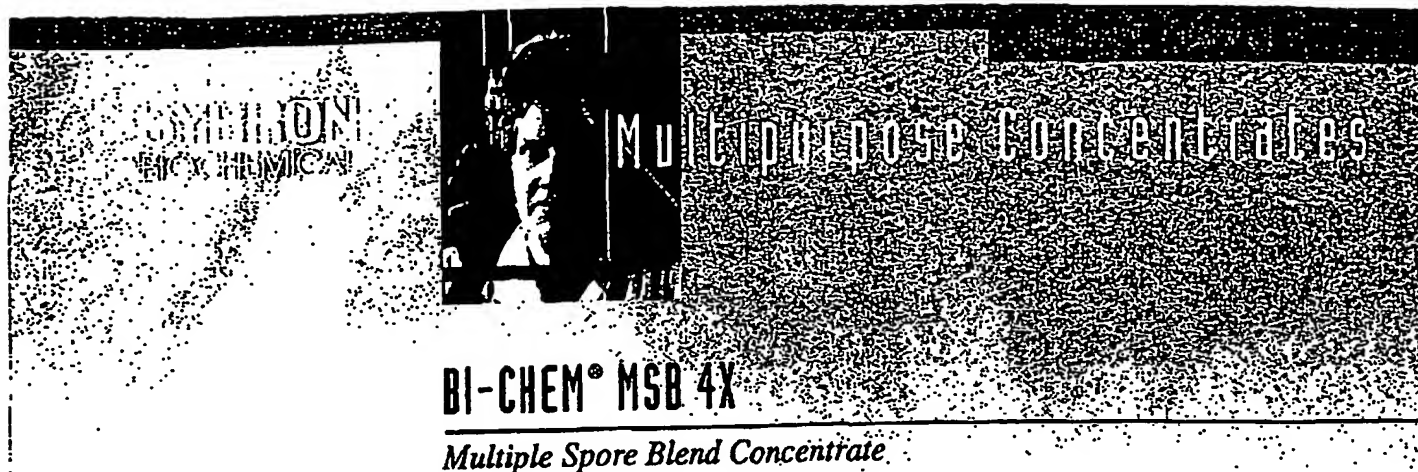
A Synergistic, Three-Strain Blend of  
Stable *Bacillus* Spore-forming Microorganisms

SOLVING A WORLD OF PROBLEMS  
THROUGH BIOTECHNOLOGY



**Declaration by Lois Davis**

**Exhibit B**



*Single product simplicity,  
multi application flexibility*

**BI-CHEM MSB 4X** is a Multiple Spore Blend concentrate designed for use as a base culture in the formulation of biological based products. This specialized microbial blend is designed to provide exceptional performance over a wide range of organic waste related applications.

**BI-CHEM MSB 4X** is available in either a non-formulated (NF) or a fully formulated (FF) concentrate. **BI-CHEM MSB 4X FF** incorporates a novel biodegradable blend of surfactants, perfume, and opacifier with the innovative multiple spore blend.

**BI-CHEM MSB 4X** is the result of intensive R&D efforts which incorporated the latest developmental techniques in biotechnology to achieve superior degradation capabilities.

The microorganisms in **BI-CHEM MSB 4X** were selected based on each strain's superior enzymatic activity against specific substrates (protein, starch, carbohydrates, fats, and greases) and the combined synergistic value of the final culture consortium,

as demonstrated in enhanced cell growth, germination rate, and effectiveness over a wide range of organic substrates. This consortium of microorganisms was also developed to perform in both aerobic and anaerobic environments.

## APPLICATIONS

**BI-CHEM MSB 4X** is a base culture designed to provide exceptional performance across multiple applications.

- *Drain Line & Grease Trap Treatment*
- *Bathroom Cleaner & Deodorizer*
- *Carpet & Fabric Care*
- *Laundry Pre-Spotter*
- *Portable Toilet Treatment*
- *Septic & Waste Treatment*

## BI-CHEM® MSB 4X

### Multiple Spore Blend Concentrate

#### PERFORMANCE

- **Enhanced Aerobic and Anaerobic Performance**, ideal for applications subject to aerobic and anaerobic environments.
- **Accelerated Enzymatic Degradation** allows the multiple spore blend to work faster and more effectively.
- **Grease Biodegradation Outperforms** other competitive formulations in laboratory testing and field studies.
- **Superior Germination and Outgrowth** results in increased bacterial activity in a variety of organic waste applications.
- **General Organic Waste Degradation**

#### CHARACTERISTICS, HANDLING, & STORAGE

##### Product Characteristics:

Bacteria Count:	21.6 x 10 <sup>9</sup> C.F.U./ml (800 Billion/gallon)
Bacterial Type:	Blend of 4 Bacillus Spores
Salmonella/Shigella:	Negative
Appearance:	
BI-CHEM MSB 4X NF:	Tan Liquid
BI-CHEM MSB 4X FF:	Creamy White
Fragrance:	
BI-CHEM MSB 4X NF:	None
BI-CHEM MSB 4X FF:	Pleasantly Perfumed
Stability:	2 years + at 35°F to 95°F
USDA Approved	

The data included herein are based on test information obtained by Sybron Chemicals, Inc. These data are believed to be reliable, but do not imply warranty or performance guarantee. We recommend that the user determine performance through laboratory testing. We assume no liability or responsibility for patent infringement resulting from the use of this product. BI-CHEM® is a registered trade mark of Sybron Chemicals, Inc.

##### Performance Characteristics:

Enzyme Production:	Lipase/Protease/ Amylase/Cellulase
Bacterial Pathways:	Aerobic and Facultative Anaerobic
pH Range:	5.0 - 9.8
Temperature Range:	38°F - 145°F

##### Storage and Handling:

Store in a cool, dry place.

Avoid inhalation, wash hands after contact, and avoid eye contact.

##### Formulation Recommendations:

Use entire content of drum.

**BI-CHEM MSB 4X NF:** Use 25% product (3 to 1 mix). Add water & chemical ingredients, mix well.

**BI-CHEM MSB 4X FF:** Use 25% product (3 to 1 mix). Add water and mix well.

##### Available Packaging:

- 5 gallon pails
- 55 gallon drums
- Bulk containers

## SYBRON BIOCHEMICAL

##### Sales offices in:

Poissy, France  
Yokohama, Japan  
Mexico City, Mexico  
Toronto, Canada

##### Ordering Information

SYBRON CHEMICAL INC.  
BIOCHEMICAL DIVISION  
Corporate Sales Office  
Birmingham Road, PO Box 66  
Birmingham, NJ 08011

U.S. Orders: (800) BUGS-HELP  
(800-284-7435)

Phone: (800) 678-0020 or (609) 893-1100

Fax: (609) 894-8641

Internet: [www.sybronchemicals.com](http://www.sybronchemicals.com)

**Declaration by Lois Davis**

**Exhibit C**



## BI-CHEM® BIOCLEAN

*A Bioenzymatic Carpet and Fabric Cleaner*

### *Effective Cleaning and Degradation of Carpet Trapped Organics*

**BI-CHEM BIOCLEAN** is designed specifically to provide a solution for the difficult removal of organics from carpet and fabric surfaces.

**BI-CHEM BIOCLEAN** provides a resolution for a broad range of stain and odor problems from the smallest spot cleaning applications to full scale cleaning applications.

**BI-CHEM BIOCLEAN** formulation is effective in the removal of organics from carpets, upholstery, draperies, car seats, etc. **BI-CHEM BIOCLEAN** will remove both old and new odors, stains and soils resulting from combined organic and inorganic soils.

#### *Spot Cleaning*

**BI-CHEM BIOCLEAN** is specifically formulated as a spot area cleaner. This is a single formulation which combines selected surfactants, perfumes, and synergistic microorganisms to achieve complete organic removal in a single cleaning application.

**BI-CHEM BIOCLEAN**, a consortium of highly active bacterial spores, was tested for the ability to degrade

organics in carpet fibers. Organic deposits in carpets result from common stains, spills and pet accidents. These organic spills lead to stains and odors. **BI-CHEM BIOCLEAN** effectively degrades these organics resulting in odor control and stain removal.

**BI-CHEM BIOCLEAN** was tested using industry standard carpet swatches with organic compounds commonly found in carpet spills. The carpet swatch inoculated with **BI-CHEM BIOCLEAN** in laboratory conditions demonstrated a 40 fold increase in degradation of organic material compared to untreated carpet.

**BI-CHEM BIOCLEAN** consists of a synergistic blend of nonpathogenic bacteria selected for their ability to degrade organic substrates. The organic spill activates the bacterial strains in **BI-CHEM BIOCLEAN**. In the presence of moisture, organic material causes the germination and growth of the microorganisms. Once the organics have been removed, the spores become dormant. The spores can then be vacuumed.

**BI-CHEM BIOCLEAN** brings nature's secret indoors for a fresher, cleaner environment by eliminating residual organics that lead to malodors.

*The natural solution to eliminate odors and organic stains.*





# BI-CHEM® BIOCLEAN

## A Bioenzymatic Carpet and Fabric Cleaner

### PERFORMANCE BENEFITS

- **General Organic Waste Degradation**
- **Enhanced Aerobic and Anaerobic Performance**, ideal for applications subject to aerobic and anaerobic environments.
- **Accelerated Enzymatic Degradation**, allows the multiple spore blend to work faster and more effectively.
- **Grease Biodegradation Outperforms** other competitive formulations in laboratory and field studies.
- **Superior Germination and Outgrowth** results in increased bacterial activity in a variety of organic waste applications.

### CHARACTERISTICS, HANDLING, & STORAGE

#### Product Characteristics:

Bacteria Count:	5.4 x 10 <sup>7</sup> C.F.U./ml (200 Billion/gallon)
Bacteria Type:	Blend of Bacillus Spores
Salmonella/Shigella:	Negative
Appearance:	Creamy White
Fragrance:	Pleasantly Perfumed
Stability	2 years + at 35°F to 95°F

#### Performance Characteristics:

High Enzyme Production:	Lipase/Protease/Amylase/ Cellulase/Urease
Bacterial Pathways:	Aerobic and Facultative Anaerobic
Optimum pH range:	6.8 - 7.0

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6/97-LMLD

#### Directions:

1. Test for colorfastness on section which does not show.
2. Remove excess solids or liquids.
3. Spray, mop or wipe a generous supply of **BI-CHEM BIOCLEAN** on to the surface to be cleaned. Following this application, use **BI-CHEM BIOCLEAN** as though it were a mild detergent to assist in the removal of the contaminants in the fabric or carpet surface.
4. Scrub any stains with **BI-CHEM BIOCLEAN**.
5. Rinse the area with fresh, warm water utilizing a sponge or soft towel in efforts to remove any residual organics.
6. In extreme cases, areas subjected to repeated soiling may require additional wetting of the surface to extend the bioenzymatic cleaning time. Place a clean wet towel over the stain for 24 hours. **BI-CHEM BIOCLEAN** provides instant odor control and contact surface cleaning, combined with bioenzymatic digestion of trapped residual organics.

#### Storage and Handling:

Store in cool, dry place.  
Avoid inhalation, wash hands after contact and avoid eye contact.

#### Available Packaging:

- 1 gallon jugs
- 5 gallon pails
- 55 gallon drums

## SYBRON BIOCHEMICAL

#### Sales offices in:

Poissy, France  
Yokohama, Japan  
Mexico City, Mexico  
Toronto, Canada

#### Ordering Information

SYBRON CHEMICALS INC.  
BIOCHEMICAL DIVISION  
Corporate Sales Office  
Birmingham Road, PO Box 66  
Birmingham, NJ 08011

U.S. Orders: (800) BUGS-HELP  
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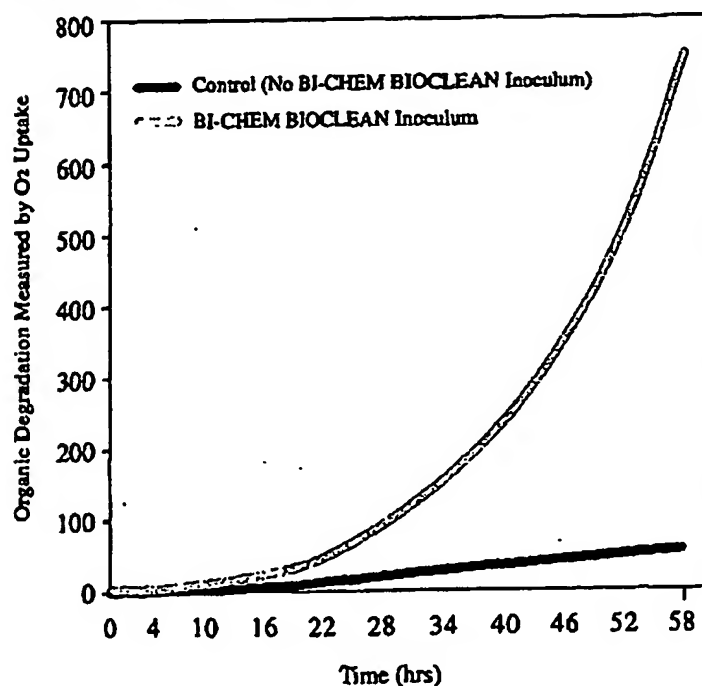
## BI-CHEM® BIOCLEAR

*Effective Cleaning and Degradation of Carpet Trapped Organics*

**BI-CHEM BIOCLEAR**, a proprietary formulation of surfactant based cleaners with a consortium of highly active bacterial spores, effectively degrades residual organics on carpet fibers. Organic deposits in carpets result from common stains, spills, and pet accidents. These deposits lead to stains and odors. **BI-CHEM BIOCLEAR** effectively degrades organics resulting in odor control and stain removal.

**BI-CHEM BIOCLEAR** was tested using industry standard carpet swatches with organic compounds commonly found in carpet spills. The carpet swatch inoculated with **BI-CHEM BIOCLEAR** in laboratory conditions demonstrated a 40 fold increase in degradation of organic material compared to untreated carpet. The graph below indicates germination and growth of spores on a carpet with spilled organics.

### BI-CHEM BIOCLEAR Outgrowth on Carpet



## BI-CHEM® BIOCLEAN

### *Effective Cleaning and Degradation of Carpet Trapped Organics*

The following electron microscopy photographs demonstrate the inoculation of bacteria on carpet fibers, which degrade residual organic spills in carpet fibers, by BI-CHEM BIOCLEAN selectively adapted bacteria.



BI-CHEM BIOCLEAN consists of a synergistic blend of non-pathogenic bacteria selected for their ability to degrade organic substrates. The organic spill activates the bacterial strains in BI-CHEM BIOCLEAN. In the presence of moisture, organic material causes the germination and growth of the microorganisms. Once the organics have been removed, the spores become dormant. The spores will then be vacuumed and removed during normal carpet maintenance.

BI-CHEM BIOCLEAN brings nature's secret indoors for a fresher, cleaner environment by eliminating residual organics that lead to malodors.

*The natural solution to eliminate odors and organic stains.*

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697-LMLD

## SYBRON BIOCHEMICAL

*Sales offices in:*

Poissy, France  
Yokohama, Japan  
Mexico City, Mexico  
Toronto, Canada

### *Ordering Information*

SYBRON CHEMICALS INC.  
BIOCHEMICAL DIVISION  
Corporate Sales Office  
Birmingham Road, PO Box 66  
Birmingham, NJ 08011

U.S. Orders: (800) BUGS-HELP  
(800-284-7435)

Phone: (800) 678-0020 or (609) 893-1100

Fax: (609) 894-8641

Internet: [www.sybronchemicals.com](http://www.sybronchemicals.com)

# **Declaration by Lois Davis**

## **Exhibit D**

# SYBRON CHEMICALS INC.

TX 25247  
 DALLAS  
 COBB CARPET SUPPLY  
 1314 VICEROY DRIVE  
 DALLAS

BIRMINGHAM ROAD, P.O. BOX 66  
 BIRMINGHAM, NEW JERSEY 08011  
 (609) 893-1100  
 FAX: 609-893-2063 ACCOUNTING  
 FAX: 609-894-8641 MARKETING  
 TELEX: 6851227

INVOICE NO	46382	INVOICE DATE	2/05/91	SHIPMENT DATE	2/04/91	SALESMAN	00797	SALES FROM	SALESMAN	NET 45	TERMS	STOCK LOCATION	SALESMAN	ORDER NO	95
NO NUMBER	210	PRODUCT NUMBER	9534E	QTY ORDERED	55	QTY SHIPPED	55	QTY	61	21-CHEM 600L EP (4X) - 55 GL	UNIT PRICE	UNIT PRICE	UNIT PRICE	UNIT PRICE	UNIT PRICE
<p>PLEASE NOTE OUR NEW "REMIT TO" ADDRESS PRINTED BELOW</p> <p>PAYMENT DUE 30 DAYS FROM SHIPMENT DATE</p> <p>SALES-TOTAL                  STATE TAX                  LOCAL TAX                  OTHER TAX                  INVOICE TOTAL</p>															

OBJECT TO TERMS AND CONDITIONS PRINTED HEREON AND ON THE REVERSE SIDE HEREOF, IF TERMS AND CONDITIONS ARE UNACCEPTABLE TO YOU, YOU MUST NOTIFY IN WRITING WITHIN 10 DAYS AS SET FORTH ON THE REVERSE SIDE OR, YOU ACCEPT OUR TERMS AND CONDITIONS.

REMIT TO ADDRESS SHOWN

P.O. BOX 640443  
 PITTSBURGH, PA 15264-0443

REMIT TO ADDRESS SHOWN

Cost allocation code #

Invoice #

Shipper's Agent

Prepared by

Requirements of the charges on the property described herein.

**Declaration by Lois Davis**

**Exhibit E**

CARPET SUPPLY  
VICEROY DRIVE

DALLAS

TX 25247

SYBRON CHEMICALS INC.

BILL TO  
COBB CARPET SUPPLY  
1314 VICEROY DRIVE

DALLAS

TX 25247

BIRMINGHAM ROAD, PO. BOX 66  
BIRMINGHAM, NEW JERSEY 08011  
(609) 893-1100  
FAX: 609-893-2063 ACCOUNTING  
FAX: 609-894-8641 MARKETING  
TELEX: 6851227

INVOICE NO.	PO	INVOICE DATE	SHIPMENT DATE	SHIPMENT FROM	TERMS	STOCK LOCATION	ORDER NO.
52978	1	5/15/91	5/13/91	SALEM, VA	NET 45	SALEM, VA	C311364
4682			00797 DENT			ROADWAY 648741063 PPD CHG	172
PO NUMBER	SALESMAN						CUSTOMER NO.
							1-91200300

QUANTITY	UNIT	DESCRIPTION	PRICE	TOTAL
9258E	SS	CL		
		PLEASE SHIP ASAP		
		BI-CHEM 600L EP (10X)NF -- SS		
		1 X SS CL DRUM		
		300 FREIGHT		
PLEASE NOTE OUR NEW "REMIT TO" ADDRESS PRINTED BELOW				
SALES TOTAL				
STATE TAX				
LOCAL TAX				
OTHER TAX				
MISC CHG-BEEN ABOVE				
INVOICE TOTAL				

PAYMENT DUE 30 DAYS FROM  
SHIPMENT DATE

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REMIT TO  
ADDRESS: PITTSBURGH, PA 15244-0043

**Declaration by Lois Davis**

**Exhibit F**



1

SHIP TO  
RUG-DOCTOR  
995 HORAN DRIVE  
FRESNO

FENTON

MO 63026

SYBRON CHEMICALS INC.

BILL TO  
RUG DOCTOR  
XACCOUNTS PAYABLE  
P O BOX 7750  
FRESNO

CA 93747

BIRMINGHAM ROAD, P.O. BOX 66  
BIRMINGHAM, NEW JERSEY 08011  
(609) 893-1100  
FAX: 609-893-2063 ACCOUNTING  
FAX: 609-894-8641 MARKETING  
TELEX: 6851227

INVOICE NO	PO	INVOICE DATE	SHIPMENT DATE	SHIPPED FROM	TERMS	STOCK LOCATION	ORDER NO.	48
84704	1	8/10/92	8/06/92	SALEM, VA	NET 45	BALEM, VA	C322270	
PO NUMBER	43050			SALESMAN	00765 WOODRUFF	SHIPPING DATA	YELLOW 2283186334 FPD CHG	CUSTOMER NO 1-91685000
PRODUCT NUMBER	QTY ORDERED	QTY SHIPPED	U/M	DESCRIPTION			UNIT PRICE	QUANTITY
				CUSTOMER F/N: 30661 MUST SHOW FOR 55 GL DRUMS AND 30671 MUST SHOW FOR 5 GAL 9/91: 55 GL -600L(10X)NF *NO SURFACTANTS/WITH EDTA USE CODE 9553E ONLY, **PLEASE SHIP TO ARRIVE ON 8-12-92** **SHIP ON THUR 8-6-92 NOT BEFORE**				
9553E	440	440	GL	HI-CHEM 600L 600L 600L 600L 8 X 55 GL DRUMS				GL
300 FREIGHT DUE 30 DAYS FROM SHIPMENT DATE								
SALES-TOTAL MISC CHG-SEE ABOVE INVOICE TOTAL								

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REMIT TO  
ADDRESS SHOWN

P.O. BOX 640443  
PITTSBURGH, PA 15264-0443

REMIT TO  
ADDRESS SHOWN

1  
1  
1

**Declaration by Lois Davis**

**Exhibit G**

**SYBRON CHEMICALS INC.**  
BIRMINGHAM ROAD, P.O. BOX 66, BIRMINGHAM, NJ 08011-0066

Telephone (609) 893-1100  
Fax # (609) 893-2063

INVOICE PAGE  
447132 1

INVOICE DATE  
11/21/94

SHIPMENT DATE  
11/18/94

**SOLD TO:**  
RUG DOCTOR  
%ACCUUNTS PAYABLE  
P O Box 7730  
FRESNO CA 93747  
US

P.O. NUMBER

SHIPPED FROM

**SHIP TO:**  
RUG DOCTOR  
997 MORAN DRIVE  
FENTON MO 63026  
US

51274

SALEM, VA

TERMS

CURRENCY

NET 45

CUST. NO.	SHIP VIA		ORDER NO.	SALESREP	
1-91685000	CFWY 46231643 FRT ALLOWED		C343376	765	BATES, JOEL
ITEM NUMBER	DESCRIPTION	U/M	QUANTITY	PRICE	AMOUNT

9533E  
DELIVER 11/29/94  
\*\*\*\*\*  
BI-13HEM 600L SPORES NS-SS  
B X SS-GAL.  
PART #30661

GL

440,000

PAY THIS AMOUNT

DIRECT TO TERMS AND CONDITIONS PRINTED HEREON AND ON THE REVERSE SIDE HEREOF. IF TERMS AND CONDITIONS ARE UNACCEPTABLE TO YOU, YOU MUST NOTIFY US IN WRITING WITHIN 10 DAYS AS SET FORTH ON THE REVERSE SIDE OR, YOU ACCEPT OUR TERMS AND CONDITIONS.  
REMIT TO  
F. N. RICH 6404433  
ACCOUNTING

**Declaration by Lois Davis**

**Exhibit H**

# SYBRON CHEMICALS INC.

BIRMINGHAM ROAD, P.O. BOX 66, BIRMINGHAM, NJ 08011-0066

Telephone (609) 893-1100  
Fax # (609) 893-2063

INVOICE # 141589

INVOICE DATE 9/15/94  
SHIPMENT DATE 9/14/94

TO: ALKEN MURRAY CORP.  
417 CANAL ST.  
NEW YORK NY 10013  
US

To: Ester

2274

SHIP: DEHAVEN TRANSFER &  
STORAGE COMPANY  
2490 PATTERSON AVENUE  
ROANOKE VA 24016  
US

P.O. NUMBER 3421  
TERMS NET 45  
CURRENCY  
SALEM, VA

CUST. NO.	SHIP VIA	ORDER NO.	SALESREP
1-81027400	DEHAVEN TRANSFER -- PICK-UP	C342667	732 RODRIGO

ITEM NUMBER	DESCRIPTION	U/M	QUANTITY	PRICE	AMOUNT
1	*****ASAP!!!! PLEASE RUSH!!!! 7/94/BCM:(0A-CUST. SPEC.- FAX & MAIL-2490 PATTERSON AVE. SW, RKE, VA (344-3643) 1/93: ALL: FAX C OF, A & COPY OF ACKNOWLEDGMENT TO BETTE 344-3643 7/94/BCM: LIQUID: ALL: DO NOT SEND IF >12 WEEKS OLD UNLESS APPROVED BY CUST. BI-CHEM 600L EP NC/NP - 5 GL 100 X 5-GL. ALKEN ENZ-ODOR	GL	500.000		

PAY THIS AMOUNT

SUBJECT TO TERMS AND CONDITIONS PRINTED HEREON AND ON THE REVERSE SIDE HEREOF. IF TERMS AND CONDITIONS ARE UNACCEPTABLE TO YOU, YOU MUST NOTIFY US IN WRITING WITHIN 10 DAYS AS SET FORTH ON THE REVERSE SIDE OR, YOU ACCEPT OUR TERMS AND CONDITIONS.

REMIT TO P.O. BOX 640443  
ADDRESS SHOWN - PATTEN BIRCHM. P.O. 15264A-0443

ACCOUNTING

# **Declaration by Lois Davis**

## **Exhibit I**

# SYBRON CHEMICALS INC.

DIKENHAY ROAD, P.O. BOX 66, DIKENHAY, NJ 08011-0066  
Telephone (609) 893-1100 Fax (609) 893-2003

AURALUX<sup>TM</sup>  
BI-CHEM<sup>®</sup>  
IONAC<sup>®</sup>  
JERSEY STATE<sup>TM</sup>  
TANATEX<sup>®</sup>

INVOICE PAGE  
488903 1

INVOICE  
DATE 4/09/98

SHIPMENT  
DATE 4/08/98

SOLD TO: QUALITECH SYSTEMS INC.  
P.O. BOX 3806  
NEW HYDE PARK NY 11040-0802  
US

SHIP TO: QUALITECH SYSTEMS INC.  
C/O SELF STORAGE CENTER  
1890 JENRICH TURNPIKE  
NEW HYDE PARK NY 11040-0802  
US

P.O. NUMBER

VERBAL-MARTIN

SHIPPED FROM

TERMS

CURRENCY

NET 45

CUST. NO.	SHIP VIA	ORDER NO.	SALESREP
-----------	----------	-----------	----------

1-9: 653400 APA PFDACHC G185526-7 C03381360 736 PATTERSON

ITEM NUMBER	DESCRIPTION	U/M	QUANTITY	PRICE	AMOUNT
2071A	BI-CHEM TD 500L - 5 GL 12 X 5 GAL PAILS; LABEL AS: ELIMINATOR II GL	GL	60.000	5.10 9.60	
2230A	BI-CHEM BIOCLEAN 12 X 5 GAL PAILS; LABEL AS: ELIMINATOR MP	GL	60.000	4.68 9.60	
	509 SHIPPING CHARGE				
	TOTAL MISC.				
	PAY THIS AMOUNT				

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REMIT TO: F.O. BOX 740039 ATLANTA, GA 30374  
ADDRESS SHOWN

**Declaration by Lois Davis**

**Exhibit J**





**Declaration by Lois Davis**

**Exhibit K**

05/25/2004 08:14 FAX 919 494 3421

NZNA LEGAL DEPARTMENT

0003

05/25/04 TUE 17:00 FAX 1 540 389 2688

NOVOZYMES BIOLOGICALS

000

COBB CARPET

PAGE 01/01

05/25/2004 14:18 2145342648

# COBB CARPET SUPPLY

Since 1962... Over 5,000 Quality Items Stocked

1314 Viceroy Drive - Dallas, Texas 75247-3911

214-634-2622 800-634-2622 Fax 214-634-2648 Web Site: <http://ticnet.users.com/cobb>

Lois Davis  
I & H Global Business Manager  
Novozymes

Dear Lois ;

We have researched our purchasing records and show original purchase orders for your Enzyme products for carpet cleaning applications. They date back to August of 1992.

It was used in our Dynachem Enzyme Deodorant for application on carpets and sold in the southern region of the United States. I have dated formulation records showing your GC-600L 10X being used as of 9/8/93.

Please give me a call if you have any more questions.

*Lawrence Cobb*

Lawrence Cobb  
Dynachem Chemical Division  
Cobb Carpet Supply



**DRIEAZ**

**DYNACHEM**

**400**



**Declaration by Lois Davis**

**Exhibit L**



**Spartan Chemical Company, Inc.**  
Manufacturer of Guaranteed Specialty Maintenance Products

May 26, 2004

Ms. Lois Davis  
Novozymes Biologicals, Inc.  
111 Kesler Mill Road  
Salem, VA 24153

**FACSIMILE TRANSMITTAL: 540/389-2688**

Dear Lois,

Brian contacted me yesterday concerning determining when exactly Spartan Chemical Company, Inc. commercialized biological based products for use in carpet cleaning and odor control. After going back through our sales records, here is what I came up with:

Spartan officially commercialized our product Consume in April of 1992. As you know, Consume was originally based on then Sybron's Bi-Chem 600 material which was composed of three different Bacillus species bacteria. Since its introduction, Consume's bacterial content and quantity have gone through numerous transitions, but the foundation of the product has remained the same. This product was developed for many different purposes, but has always been labeled with a directional heading of "CARPET ODOR AND STAIN REMOVER".

[REDACTED]

I hope this covers what you need Lois. If we can provide any further information or documentation in this matter please do not hesitate to contact me. We greatly appreciate your efforts!

Sincerely,

SPARTAN CHEMICAL COMPANY, INC.

*William J. Schallitz*

William J. Schallitz  
Vice President, Research & Development



Corporate Administration  
1110 Spartan Drive, Maumee, OH 43537  
Phone: 419-331-5551 / Fax: 419-336-8429  
Toll Free: 1-800-537-8990



Manufacturing/Customer Service/Orders  
1110 Spartan Drive, Maumee, OH 43537  
Phone: 419-897-5551 / Fax: 419-897-9262  
Toll Free: 1-800-537-8990

Toll-Free Fax On-Demand: 888-923-4858  
Electronic Bulletin Board: 419-897-0859  
Web Site: [www.spartanchemical.com](http://www.spartanchemical.com)  
E-Mail: [spartan@spartanchemical.com](mailto:spartan@spartanchemical.com)

**Declaration by Lois Davis**

**Exhibit M**

AVERAGE UNIT PRICE  
MONTH YTD PR.YTD

DATE 04/02/77 ORDER BY ELLIOTTAN BY CUSTOMER BY PRODUCT AS OF 04/97 MONTH D.O.L L.A.R 9 PR.YTD

PRODUCT NAME	MONTH	YTD	PR.YTD
BI-CHEM MANTA-10000	55	140	
BI-CHEM MANTA-10000	55	165	
BI-CHEM MANTA-10000	55	75	
BI-CHEM MANTA-10000	55	75	
BI-CHEM MANTA-10000	55	40	
BI-CHEM MANTA-10000	55	110	
TOTAL	225	405	
BI-CHEM MANTA-10000	2000	200	
BI-CHEM MANTA-10000	2000	550	
BI-CHEM MANTA-10000	2000	2000	
TOTAL	2000	2750	
BI-CHEM MANTA-10000	110	220	
TOTAL	110	220	
BI-CHEM MANTA-10000	5400	5400	
BI-CHEM MANTA-10000	1300	1300	
BI-CHEM MANTA-10000	250	250	
BI-CHEM MANTA-10000	375	375	
BI-CHEM MANTA-10000	330	330	
BI-CHEM MANTA-10000	1705	1705	
TOTAL	9505	9505	
BI-CHEM MANTA-10000	110	110	
TOTAL	110	110	
BI-CHEM MANTA-10000	50	50	
TOTAL	50	50	
BI-CHEM MANTA-10000	55	55	
TOTAL	55	55	
BI-CHEM MANTA-10000	1600	1600	
TOTAL	1600	1600	
BI-CHEM MANTA-10000	55	55	
BI-CHEM MANTA-10000	1600	1600	
BI-CHEM MANTA-10000	440	440	
BI-CHEM MANTA-10000	2095	2095	
TOTAL	1650	1650	
BI-CHEM MANTA-10000	1375	1375	
TOTAL	3025	3025	

M

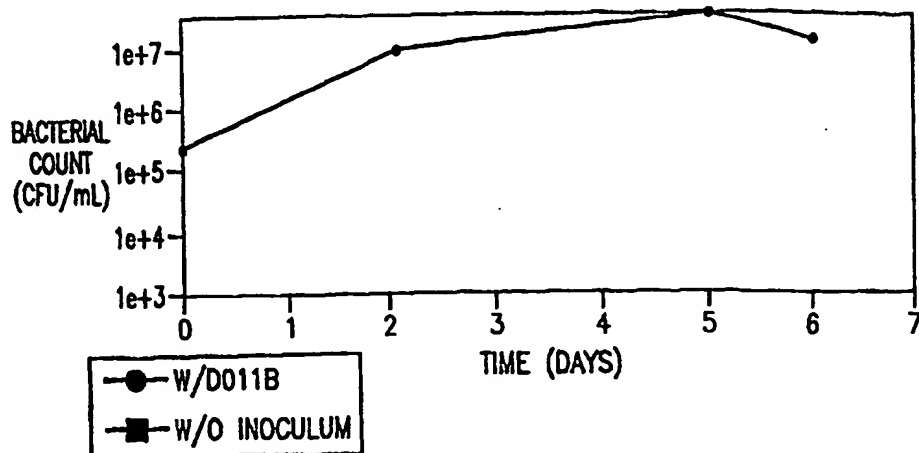
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## INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification <sup>6</sup> : C09K 3/00, A01N 43/08, A61K 31/34		A1	(11) International Publication Number: WO 99/46350
			(43) International Publication Date: 16 September 1999 (16.09.99)
(21) International Application Number: PCT/US99/04812		(81) Designated States: AU, BR, CA, CN, IL, JP, KP, KR, MX, NO, NZ, SG, European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE).	
(22) International Filing Date: 5 March 1999 (05.03.99)			
(30) Priority Data: 60/077,544 11 March 1998 (11.03.98) US		Published With international search report.	
(71) Applicant: SYBRON CHEMICAL HOLDINGS, INC. [US/US]; Suite 1300, 1105 North Market Street, Wilmington, DE 19899 (US).			
(72) Inventors: LIN, Jian-Er; 6107 Scotford Court, Roanoke, VA 24018 (US). McPEAK, John, F.; 395 Kings Highway, Nickleton, NJ 08056 (US).			
(74) Agent: WALL, Thomas, J.; Wall Marjama Bilinski & Burr, Suite 400, 101 South Salina Street, Syracuse, NY 13202 (US).			

(54) Title: BIOLOGICAL DEODORIZING LIQUID COMPOSITION



(57) Abstract

The composition of the invention comprises an aqueous mixture of an odor neutralizer component, an enhancer component for microbial activity, and a microbial component. This composition is designed to provide short- and long-term odor control effects and is environmentally friendly and economical for use.



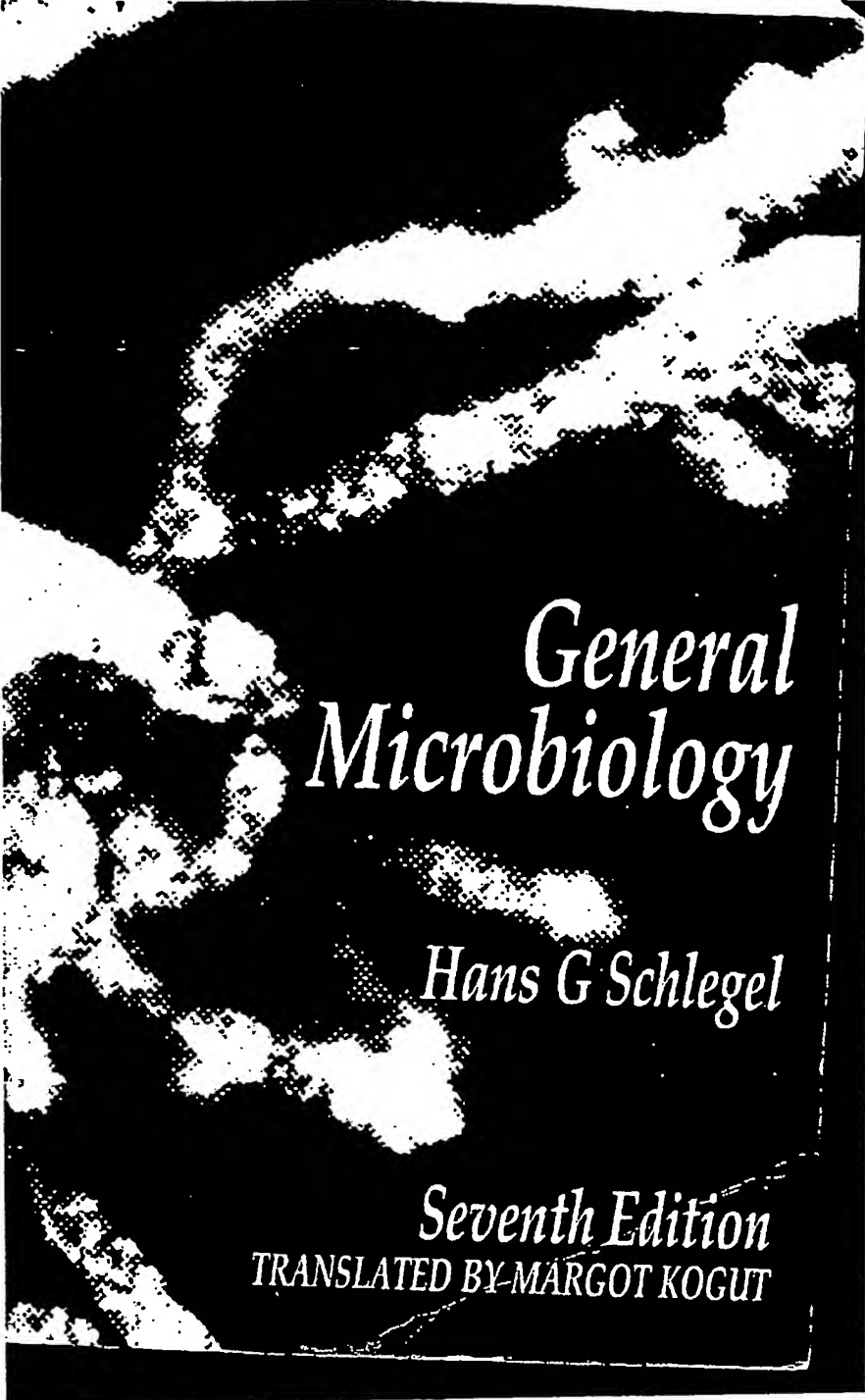


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(51) International Patent Classification 6 : A01N 63/00, 63/04, C11D 3/38, 3/00	A1	(11) International Publication Number: WO 97/25865
		(43) International Publication Date: 24 July 1997 (24.07.97)
(21) International Application Number: PCT/US97/00380 (22) International Filing Date: 10 January 1997 (10.01.97)  (30) Priority Data: 08/585,912          16 January 1996 (16.01.96)          US  (71) Applicant: SYBRON CHEMICAL HOLDINGS, INC. [US/US]; Suite 1300, 1105 North Market Street, Wilmington, DE 19899 (US).  (72) Inventors: LIN, Jian-Er; 6107 Scotford Court, Roanoke, VA 24018 (US). DENT, Douglas, A.; 10 Candle Wyck Court, New Hope, PA 18938 (US).  (74) Agent: MARJAMA, Owen, D.; Harris Beach & Wilcox, L.L.P., One Park Place, 4th floor, 300 South State Street, Syracuse, NY 13202 (US).		(81) Designated States: AU, BR, CA, CN, JP, KP, KR, MX, NO, SG, European patent (AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE).  Published <i>With international search report.          Before the expiration of the time limit for amending the claims and to be republished in the event of the receipt of amendments.</i>
(54) Title: CLEANER AND SANITIZER FORMULATION  (57) Abstract  The formulations of the invention comprise a suspension of a sanitizing composition, bacterial spores, surfactants, a thickening agent, and abrasive particles all contained in aqueous solution. These formulations can be used for cleaning and sanitizing bathroom fixtures, sinks, toilet bowls, and other dirty and contaminated surfaces, and have the advantages of being a good surface cleaning agent and a good sanitizer, along with providing the long-term effect of beneficial bacteria that control pathogens and degrade wastes both on the surface and in the sewage system associated with the surface being treated.		



# *General Microbiology*

*Hans G Schlegel*

*Seventh Edition*  
TRANSLATED BY MARGOT KOGUT

Published by the Press Syndicate of the University of Cambridge  
The Pitt Building, Trumpington Street, Cambridge CB2 1RP  
40 West 20th Street, New York, NY 10011-4211, USA  
10 Stamford Road, Oakleigh, Melbourne 3166, Australia

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Second edition 1993

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QR41.2.S3413 1992

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ISBN 0 521 43372 X hardback

ISBN 0 521 43980 9 paperback

KS

suspension in water is no longer (completely) effective in this repression; the induction (or derepression) of sporulation persists and between the tenth and thirteenth hour after transfer to water, about 90% of the cells have sporulated. It appears, therefore, that spore formation is regulated by environmental factors. The yield of sporulating cells can often be increased by the addition of manganese salts to the medium.

The capacity to form endospores can be lost gradually by repeated subculturing of vegetative cells (under non-sporulating conditions). Since suspensions of sporogenous organisms contain vegetative cells as well as spores (under appropriate conditions), they are usually briefly exposed to boiling water temperatures before subculturing. This treatment helps to maintain or even increases the capacity of the culture to form endospores.

**Properties of mature spores.** Spores are liberated upon autolysis of the vegetative cell. The mature spores have no demonstrable metabolic activity and exhibit a high degree of resistance to heat, radiation, and chemicals. Their heat resistance is due to the low water content of spores. *Bacillus megaterium* spores, for example, have a water content of 15%, similar to that of wool or dry casein. Lyophilised vegetative bacteria are also very heat resistant. In addition, the heat resistance of spores appears to be proportional to their dipicolinic acid content. The radiation resistance of spores is also greater than that of vegetative cells, and it is approximately proportional to the number of disulphide bridges present in the outer protein layers. The spore envelope mainly contains a cysteine-rich protein which resembles keratin. The chemical resistance of spores is due to the impermeability of the spore envelope to many chemicals.

**Spore germination.** In suitable nutrient media, most spores can be induced to germinate. However, certain pretreatments, such as storage and brief exposure to high temperature, can increase the percentage of germination of a spore population. For *Bacillus subtilis* spores, for instance, a 'rest period' of 7 days and heating for 5 minutes at 60 °C are regarded as optimal conditions for germination. Other spores can be 'activated' by exposing to boiling water temperature (100 °C) for 10 minutes. This heat-shock treatment must be carried out immediately before placing the spores on germination medium, since the activation process(es) appear to be reversible. Germination of spores is preceded by water uptake and swelling of the spores. In some cases it is dependent on the presence of glucose, amino acids, nucleosides, or other substances. During germination, fundamental physiological alterations take place: respiration and enzymatic activities increase rapidly; amino acids, dipicolinic acid, and peptides are excreted; and about 25–30% of the dry

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